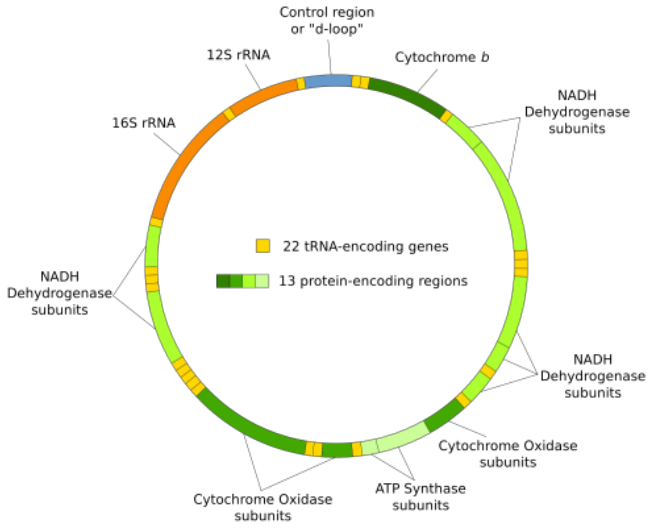
Mitochondrial DNA mutations: The good, the bad, and the ugly

13 January 2015, by John Hewitt



Mitochondrial DNA. Credit: en.wikipedia.org

(Medical Xpress)—Programmers typically evolve new code by copying and modifying existing code to meet new needs. With the more advanced programming languages, they also make use of something known in the business as polymorphism—the ability to process objects differently depending on their data type or class. Similarly, one way that life evolves is to copy and modify genes. Biologists, however, often use the term polymorphism to mean different things. Sometimes it simply means a non disease-causing change to a base pair, and sometimes it more specifically means a change found at a frequency of 1% or higher in the population.

The word 'mutation' is often associated with something negative, a disease causing variant or a pathogenic subsitution. The problem with these kinds of terms is that despite their different meanings, they can and will be used to describe the exact same change in any number of specific base pair alterations. This is no way to run scientific dialog, let alone research. At the risk of getting bogged down in such ambiguities, we might suggest that the central question of what is the meaning of *change* in base pairs can be still answered, only not yet for our nuclear DNA. On the other hand, our rapidly mutating mitochondrial DNA (mtDNA) has just 37 genes and therefore provides a much simpler and compact microcosm to explore. Of those genes, 22 encode already familiar tRNAs, two are for rRNAs, and the remaining 13 spots code subunits for just 4 proteins—ATP synthase, NADH dehydrogenase, cytochrome oxidase, and cytochrome b.

A recent review in *Neuron* provides a nice summary of the current state of affairs for mitochondrial mutations in the central and peripheral nervous system. Why the nervous system you might ask? The simple answer is that due to the high demand for oxygen in the brain, and therefore for mitochondria, this is where the effects of changes tend to show up. The more complex answer for why mitochondrial mutations have disproportionate effects on the brain leads to a possible explanation for the origins of neurons themselves: their spatially-extended structure, having been evolved largely by and for their mitochondria, provides both refuge from a potentially hostile nuclear environment where they are continually threatened with lysosomal degradation, and also a way to select and deliver mitochondrial derivatives and mtDNA nucleoids to parts unknown.

Among the more specific assaults to mtDNA, is a sniper-like ability seen in Leber's hereditary optic neuropathy (LHON) to uniquely target retinal ganglion cells. The most commonly found mutation of this type looks something like this: LHON ND4 m.11778G>A. In keeping with the official Human Genome Variation Society guide this nomenclature would indicate that this particular defect is caused by a subsitution of adenine for guanine at position 11778 in the 4th subunit of mitochondrial NADH dehydrogenase. On an even more interesting note,

these same ganglion cells are known to do something that until now would have been called unusual. Within the lamina of the optic nerve head, before the ganglion cell axons pick up a thick coat of myelin, they select specific mitochondria according to their specific markers and membrane potential, and shed them to their surroundings.

This one-two punch, transexudation followed by transmitophagy, delivers mitochondria to a surrounding meshwork of orthogonally oriented astrocytic processes that eagerly consume them. I asked Mark Ellisman and Nicholas Marsh-Armstrong, the researchers who discovered this, if these events are affected in LHONs or other mtDNA disorders. While they have not looked to deeply into this question specifically, they indicated that new work suggests these events are much more widespread in the brain then we might first imagine, and furthermore that there appears to be cyclic or circadian variation.

The brain is not the only place where mtDNA is routed between cells, or for that matter between organs. Mike Berridge just published work in the journal *Cell Metabolism* showing that tumor cells from the breast and skin fail to thrive when DNA for respiratory proteins is deleted from their mitochondria (creating so-called rho0 mitochondria). When transplanted in mice, these cells somehow later acquire mtDNA, presumably through horizontal transfer from the host. In the absence of any clear mechanism for uptake of the necessary DNA, the best explanation seemed to be that mitochondria are transferrred to the new cells.

Berridge was able to show that this is the case by comparing mtDNA polymorphism between different mouse strains and cell lines. I also asked him if he thought his results might reflect a similar process to that going on in opic nerve transmitophagy. He noted that the packaging of questionable mitochondria for astrocytic recycling is complimentary to his study and raised the question of whether astrocytes could in turn be a source of yound mitochondria for synapses, especially those at considerable distance from the cell body.

The above observations regarding mitochondria in neurons and mitochondria in tumor cells might be tied together by considering their role in the generation of oocyte and its subsequent differentiation into unique cells in the development of the early embryo. Cancer cells invariably show increased glucose utilization and decreased mitochondrial respiration. This so-called 'Warburg effect' makes them stand out in PET scans where their rapid glucose uptake provides a convenient way to diagnose and track their response to treatment. One explanation of this effect is that cancer-related genes shut down mitochondria that would normally be involved in the apoptosis program that normally kills tumor cells. Crippled or reduced numbers of mtDNA copies could also be an adaptation to the low-oxygen environment in tumor cells.

More convincing perhaps, is the idea that cancer cells shift towards glycolysis despite the presence of oxygen because glycolysis directly provides the building blocks needed for cell proliferation in the right place and proportion. A similar situation may be seen during cell proliferation in the initial stages of the developing embryo where mtDNA replication is temporarily halted for a number of cell division cycles that depends on the particular species. This comes after a phase of mitochondrial proliferation in the maturing oocyte which was preceeded by an even more dramatic mitochondrial genetic bottleneck where the number of mtDNA copies per mitochondria, and per cell was decimated. This restriction-amplification-restriction process is one way the oocyte, and the organism, selects the right mix or heteroplasmy from a pristine population of virginal mitochondrial templates to contribute to the offspring. This embryological Santa Claus, which apportions mitochondria according to an as yet fully seen mechanism may reflect much the same purification and quality control processes we noted in neurons above.

In this view, a mitochondrium effectively wears a given mtDNA variant given its sleeve—it represents the entire organelle which offers itself whole for selection in a way that a single nuclear or germline mutation, amidst a huge background of nuclear DNA variance in each generation, rarely can. But here there is another even more powerful angle of attack to consider. Many ideas in life are best understood by their exceptions, and paternal

inheritance of mitochondria is certaining exceptional. Some species of mussels, in particular, do their mitochondrial inheritance a bit differently from the rest of us, and therein lies the magic. At first glance it would appear that they violate the rule of uniparental inheritance because (at least in males), they don't block or target mitochondria donated by sperm for degradation. In most other species, the question is not if, but when and where sperm mitochondria will eventually be eliminated.

However, this apparent violation of uniparental inheritance vanishes when we observe that independent segregation is still maintained; namely, the female type mitochondria are transmitted from mothers in both sexes while the male type is only transmitted from fathers to sons. The important thing for us is that while sperm mitochondria are evenly dispersed among subsequent developing blastomeres to generate females, to make males, sperm mitochondria are concentrated specifically in a blastomere known as 4d. What this means is that not only does the developing embryo have the ability to distinguish mitochondria, but it can accurately position them as well. For example, after fertilization some sperm mitochondria are released in the area where division will later occur, and in other instances they have been observed to concentrate at the cleavage furrow at the 2-4 cell stage.

Since early embryos asynchronously split just one cell at a time, setting a universal speed limit via controlling mtDNA replication and therefore energy available to the embryo enables cells to individually be given identity. As we mentioned, every species has its own differention program, closely regulating the total number of mtDNA copies per cell, and per embryo, up to some given level. For mice that level is the 2 cell stage, and for pigs and cattle the 8 cell stage, while in others it can variously be the blastula, morula or gastrulation stage.

Many of the other specific disease-causing mutations mentioned in the review are not surprisingly found in the tRNA genes. One outstanding question is that considering the similarity of some nuclear encoded tRNAs (like met-tRNA) to mito-tRNAs, what would it take for mitochondrial to utilize nuclear tRNAs, and similarly vice-versa? In other words, do we know that this does not happen? Many of the proteins associated with nucleoids of mtDNA have been shown to be surprisingly versatile. For example, in yeast, both aconitase and Ilv5 are bifunctional with distinct activities both as metabolic proteins and in maintaining mitochondrial nucleoids. Of note, they have been linked to mtDNA positioning and segregation as well as mitochondrial translocations, particularly those associated with cell division.

Researchers currently are unable to manipulate the mitochondrial genome with the same facility that they can manipulate nuclear sequences. However, as the their ability to insert mutation reporters or selectable markers into the mitochondrial genome increases, some of the questions raised above by looking into early devlopmental process for inspiration may be better answered.

A lot of fascinating new results regarding single mitochondrial mutations and their amplified macroscopic effects on the nervous system were left out. They have names like Twinkle, Polg, MELAS, and MERRF. With any luck we can do another review called, Everything you wanted to know about mitochondrial mutations but were afraid to ask.

More information: — Mitochondrial DNA: Impacting Central and Peripheral Nervous Systems, *Neuron*, Volume 84, Issue 6, p1126–1142, 17 December 2014. dx.doi.org/10.1016/j.neuron.2014.11.022

— Mitochondrial Genome Acquisition Restores Respiratory Function and Tumorigenic Potential of Cancer Cells without Mitochondrial DNA, *Cell Metabolism*, Volume 21, Issue 1, p81–94, 6 January 2015. dx.doi.org/10.1016/j.cmet.2014.12.003